Early Embryogenesis

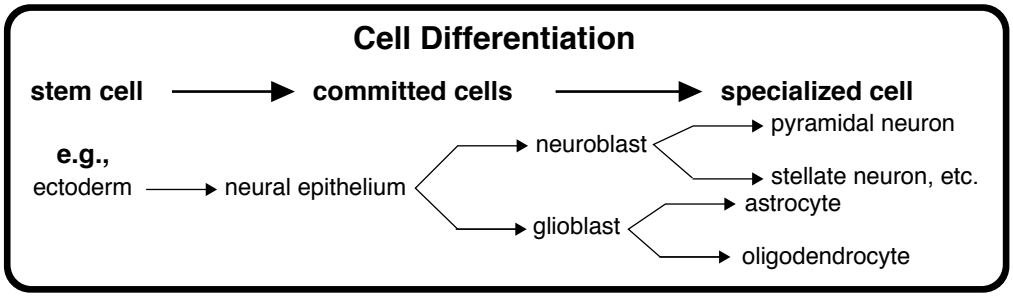
Embryogenesis:

— formation of body structures & organs (organogenesis)
— requires cell division (proliferation) and cell differentiation (specialization)
— produces the great variety of cell types and extracellular products found in the body.

Cell specialization:

— selective gene expression (and resultant protein production) is the ultimate explanation for the cell differentiation process during embryogenesis.

— genetic expression by a particular cell depends on the cell's previous genetic history (commitment lineage) and its current cellular environment (intercellular communications).



Cell differentiation is the result of cells expressing some genes and suppressing others within a common genome. Cells differ because they produced different proteins/peptides.

Proteins & peptides are:
— structural components (cytoskeleton or extracellular structures)
— enzymes (controlling cell metabolism)
— secretory products (e.g., hormones; digestive enzymes; etc.)
— channels & pumps (passage of molecules across membranes)
— receptors (communication, etc.)

Periods:

Embryonic Period — defined as the time from fertilization to the earliest (primordial) stages of organ development (about 30 days in dog, cat, sheep, pig; almost 60 days in horse, cattle, human).

Fetal Period — the time between the embryonic period and parturition (the end of gestation), during which organs grow and begin to function.

Fertilization:

— union of a haploid oocyte and a haploid spermatozoon, producing a diploid zygote (a pleuripotent cell capable of developing into a new individual)
— fertilization begins with gamete fusion (zygote formation)
— fertilization ends with the initiation of zygote cell division (the start of cleavage)

Fertilization related details:

— fusion of a spermatozoon with an oocyte takes place in the uterine tube, near the ovary
— the spermatozoon must bind to a specific glycoprotein on the zona pellucida surrounding the oocyte [this species recognition process prevents union with foreign sperm];

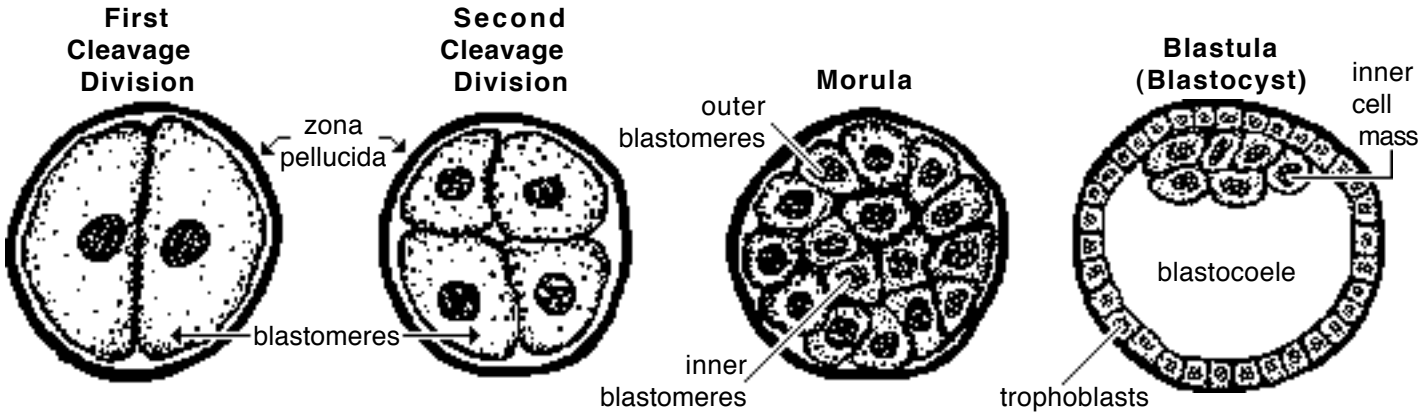
— then the spermatozoon releases degradative enzymes (acrosomal reaction) [the enzymes denature the zona pellucida, allowing the sperm cell to penetrate the barrier]
— spermatozoon and oocyte plasma membranes fuse (secondary oocyte completes meiosis)
— the oocyte immediately cancels its membrane potential (via Ca^{++} influx) and then denatures its zona pellucida (via enzymes are released by exocytosis from oocyte cytoplasmic granules) [this prevents fusion by additional sperm]
— male & female haploid pronuclei make contact, lose their nuclear membranes, and begin mitosis (mitosis begins 12 hours after sperm fusion; DNA synthesis takes place before mitosis)

Oocyte (enveloped by a zona pellucida (glycoprotein membrane) and corona radiata (granulosa cells) at ovulation)
— selective follicles mature at each cycle (in response to circulating FSH hormone from the pituitary)
— oogonia (germ cells) give rise to primary oocytes by mitosis within the embryo
— primary oocytes initiate Meiosis I (reduction division) within the embryo and only resume Meiosis I following ovulation (being suspended in Meiosis I by inhibitory secretion of follicle granulosa cells)
— secondary oocytes complete meiosis (Meiosis II) following fertilization (if unfertilized they degenerate), producing a fertilized oocyte (ovum).

Spermatozoa (several hundred million per ejaculate)
— propelled from vagina to uterine tube by contraction of female genital tract
— spermatogonia (germ cells) give rise to primary spermatocytes by mitosis repetitively following puberty
— primary spermatocytes undergo Meiosis I (reduction division) producing secondary spermatocytes
— secondary spermatocytes complete meiosis (Meiosis II), producing spermatids that undergo transformation into spermatozoa (spermiogenesis)
— subsequently, spermatozoa undergo capacitation (removal of surface proteins that would impede contact with an oocyte)

Cleavage:

— refers to the initial series of mitotic divisions by which the large zygote is fractionated into numerous "normal size" cells.
— each daughter cell of the cleavage process is termed a *blastomere*.
— cleavage begins with a *zygote*, progresses through compaction to a *morula* stage and terminates at the start of the *blastocyst* (blastula) stage
— the first eight blastomeres are undifferentiated and have identical potential in mammals; thereafter, blastomeres differentiate into inner & outer cells with different missions



Note: The first cleavage division occurs 1 to 5 days following ovulation (depending on species), thereafter cells divide about once every 12 hours;
As many as eight generations of mitoses may occur without intervening cell growth (cytoplasmic increase). Thus, e.g., one 150 micron diameter zygote can becomes a collection of 256 cells, each about 7 microns in diameter.

Morula [L.= small mulberry]

— a solid ball of blastomeres within a zona pellucida (typically consisting of 16 to 64 blastomeres)
— blastomeres become compacted; cells on the inside differentiate from those along the surface of the morula:
— *outer blastomeres* become flattened and form tight junctions (reducing fluid permeability); they develop the capacity to secrete fluid (internally); they are destined to become ***trophoblasts*** which form the chorion & amnion (fetal membranes) of the conceptus;
— *inner blastomeres* form gap junctions to maximize intercellular communication; they are destined to become ***inner cell mass*** which forms the embryo itself (plus two fetal membranes).

Note:
- As few as three inner blastomeres are sufficient to produce an entire embryo (and adult).
- When a morula leaves the uterine tube and enters the uterus (uterine horn) it is at about the 16-cell stage, around 4 to 7 days after fertilization (depending on species).
- The 32-cell stage morula (5-7 days post ovulation) is ideal for embryo transfer in cattle.

Blastocyst (or Blastula)

— develops during the second week, after the zona pellucida ruptures
— consists of a large number of blastomeres arranged to form a hollow, fluid-filled, spherical or cylindrical structure
— contains an **inner cell mass** (embryoblast), evident as a collection of cells localized inside one polar end of the blastula
— surface cells of the blastocyst are designated **trophoblasts** (future chorion of the conceptus)
— the cavity of the blastocyst is called a **blastocoele**
— eventually the blastocyst attaches to or implants within the uterine wall (pending species).

Cleavage in fish, reptiles, and birds:

Large quantities of yolk impede cell division during cleavage. Thus a *blastodisc* (rather than a spherical or elliptical *blastocyst*) is formed at the animal pole of the egg.

A telolecithal ovum (egg with large amounts of asymmetrically distributed yolk) has an *animal pole* where the nucleus is located and an opposite *vegetal pole* where yolk is concentrated. Cleavage is partial (meroblastic): cells divide more rapidly at the animal pole than at the vegetal pole, resulting in many, small blastomeres at the animal pole and a few, large macromeres at the vegetal pole.

In contrast, mammalian ovum has meager amounts of yolk (oligolecithal ovum) which is uniformly distributed (isolecithal). Cleavage is holoblastic (total) and each blastomere division produces two equal-size daughter cells. Thus animal and vegetal poles are not evident in mammalian ova.

TWINS

Monozygotic: identical (same genetic composition) twins can result from either:

1] separation of <u>early blastomeres</u> (up to the 8-cell stage)—each of the separate blastomere(s) develops into an independent conceptus; or

2] separation of <u>inner blastomeres</u> within a single morula—each of the separate blastomere(s) develops into an independent embryo and both embryos share a common placenta (this is less common than the first possibility).

Note: Separations later in embryonic development result in conjoined twins (diplopagus; Siamese twins), or double heads, etc. types of anomalies.

Dizygotic: fraternal twins result when two zygotes develop "independently" during the same pregnancy (independence can be compromised by fusion of fetal membranes and blood supplies). It is possible for fraternal blastomeres to merge and produce a single conceptus with two different genotypes (a chimera).

GERM LAYERS

Ectoderm, **mesoderm** and **endoderm** are designated *primary germ layers* because origins of all organs can be traced back to these three layers.

Ectoderm forms epidermis of the skin, epithelium of the oral and nasal cavities, and the nervous system and sense organs.

Mesoderm forms muscle and connective tissue, including bone, and components of the circulatory, urinary and genital systems.

Endoderm forms mucosal epithelium and glands of respiratory and digestive systems.

Gastrulation:

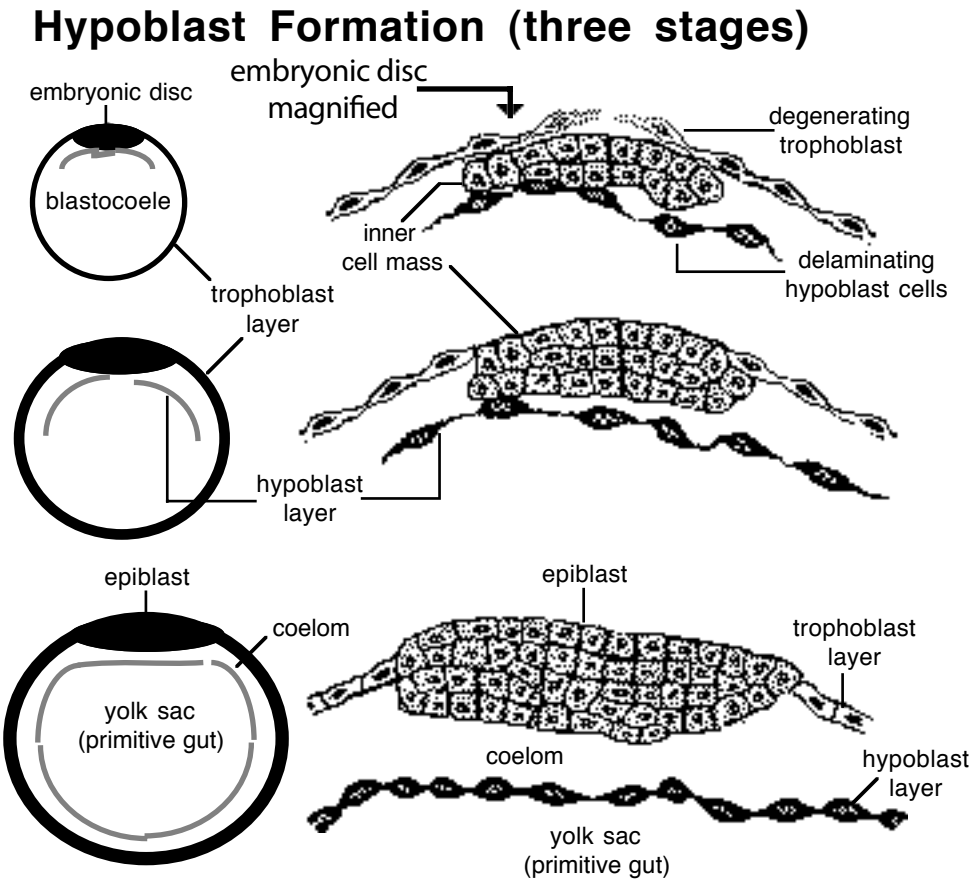
The morphogenic process that gives rise to three germ layers: **ectoderm**, **mesoderm**, and **endoderm**. (In some species, evidence of primitive gut formation can be seen [*gastrula* Gr.= little stomach] .)

Gastrulation includes the following sequence, beginning with a blastocyst:

— A thickened embryonic disc becomes evident at the blastocyst surface, due to cell proliferation of the inner cell mass cells. Trophoblast cells overlaying the inner cell mass degenerate in domestic mammals (in the mouse and human, trophoblast cells overlaying the inner cell mass separate and, instead of degenerating, become amnionic wall.)

— From the inner cell mass, cells proliferate, break loose (delaminate), and migrate to form a new cell layer inside the trophoblast layer. The new layer of cells, called the *hypoblast*, will form a yolk sac. The remaining inner cell mass may be called the *epiblast*.

— On the epiblast surface, a **primitive streak** forms as differential cell growth generates a pair of ridges separated by a depression. [NOTE: The primitive streak defines the longitudinal axis of the embryo and indicates the start of germ layer formation.]

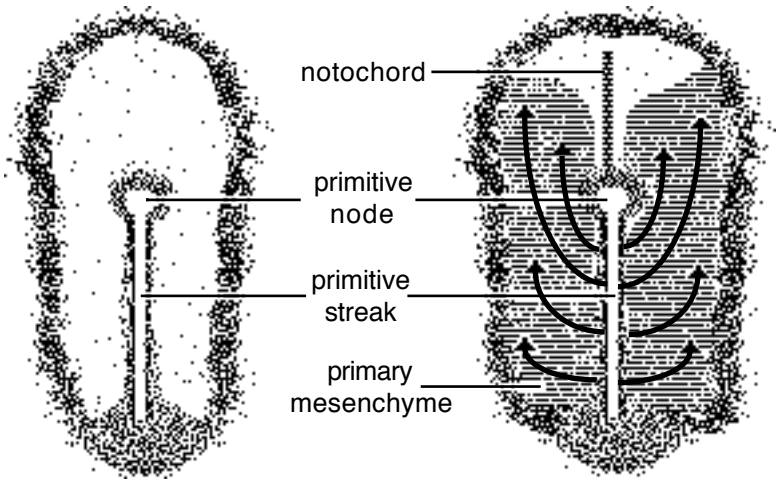


— Deep to the primitive streak, a space (**coelom/celom**) becomes evident between the hypoblast layer and epiblast. Subsequently, the coelom is filled by mesoderm that undergoes cavitation and gives rise to body cavities.

— Epiblast cells proliferate along primitive streak margins and migrate through the streak into the coelom. The migrating cells form endoderm & mesoderm layers.

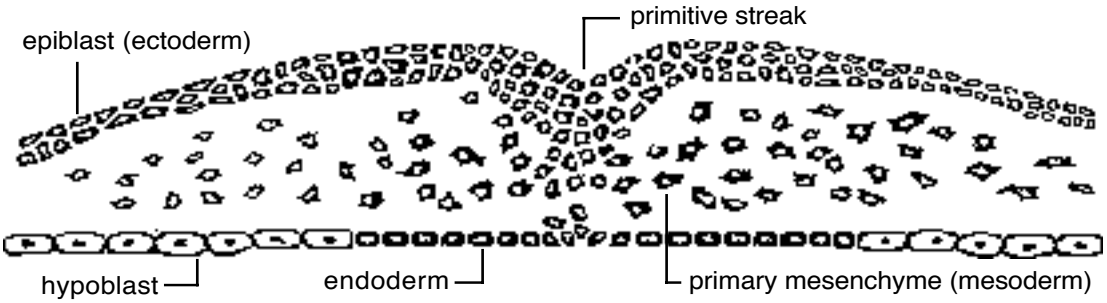
— Initial migrating cells join the hypoblast layer, forming embryonic **endoderm** (hypoblast cells constitutes yolk sac endoderm).

Dorsal View of Embryonic Disc



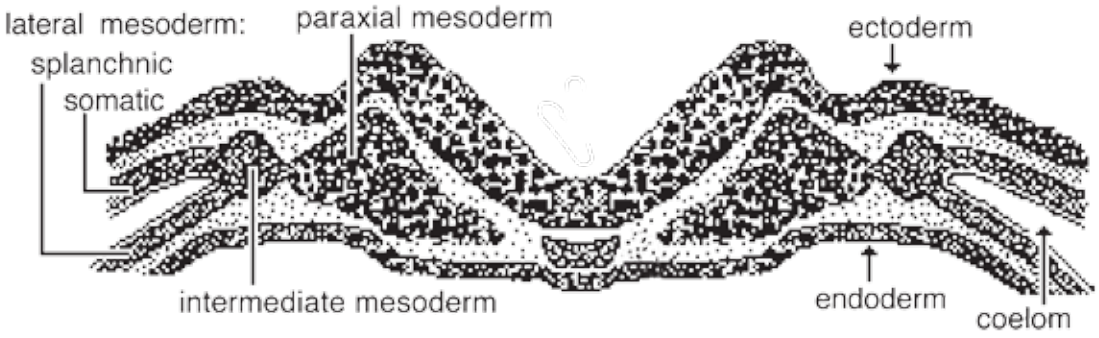
NOTE: Arrows indicate the spread of primary mesenchyme through the primitive streak and between the epiblast and hypoblast

— The majority of migrating cells enter the coelom as primary mesenchyme and become **mesoderm**. The primary mesenchyme migrates laterally and cranially (but not along the midline region directly cranial to the primitive streak where notochord will form). Note: Mesoderm divides into: paraxial, intermediate, and lateral mesodermal regions.



— Within the lateral mesoderm, cavitation re-establishes a coelom (hoseshoe-shaped). The mesoderm splits into two layers bordering the coelom—*somatic mesoderm* is attached to the ectoderm and *splanchnic mesoderm* is joined to endoderm.

— The remaining epiblast becomes **ectoderm** which forms skin epidermis & nervous system.



NOTE:

Mesoderm can exist in two morphologic forms: mesenchyme and epithelioid:

Mesenchyme features aggregates of stellate cells within an abundant extracellular matrix composed of fluid and macromolecules (polymers).

Epithelioid refers to organized cells having distinct apical and basal surfaces; the latter commonly rests on a basal lamina produced by epithelioid secretion.

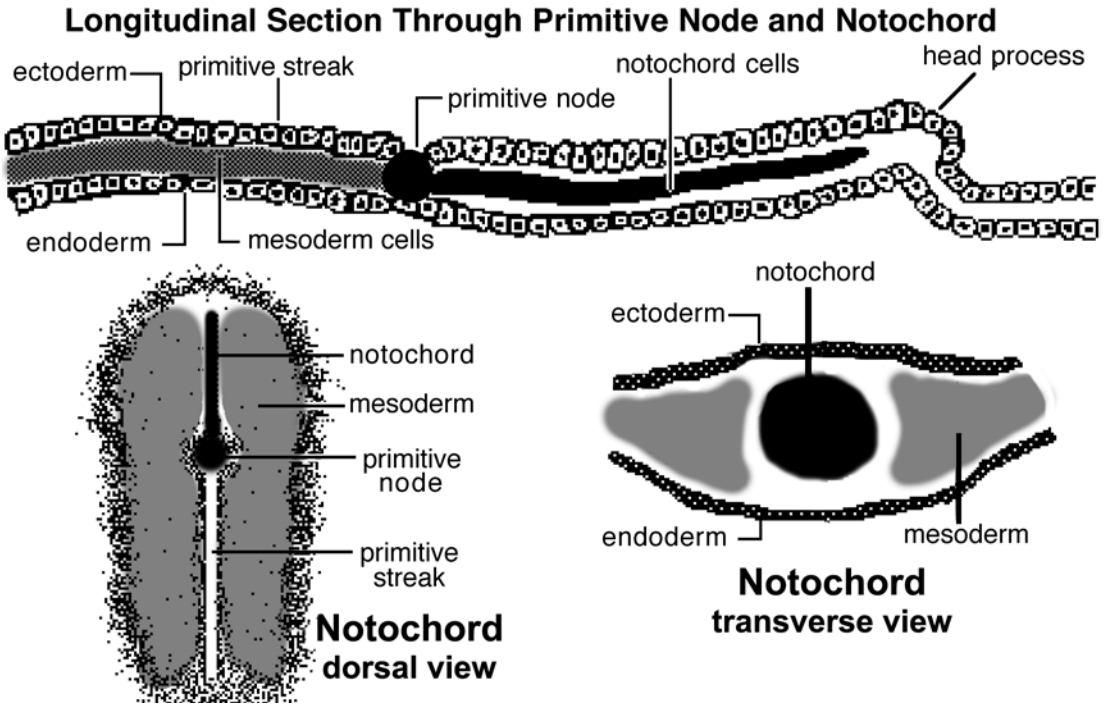
Mesoderm can transform from a mesenchyme to epithelioid and vice versa: The mesoderm that streams through the primitive streak is *primary mesenchyme*. Somatic, splanchnic, and somite mesoderm can be temporarily epithelioid. The temporary epithelioid transforms to a *secondary mesenchyme* which ultimately forms muscle and connective tissue (including cartilage, bone, ligaments, tendons, dermis, fascia, and adipose tissue).

Thus, the term "mesenchyme" refers to the morphologic appearance of embryonic tissue. Although most mesenchyme is mesoderm, the other germ layers can also form mesenchyme, e.g., ectomesenchyme from neural crest ectoderm.

Formation of the Notochord:

- The **notochord** is a rod-shaped aggregate of cells located between ectoderm and endoderm anterior to the primitive streak of the embryo. It occupies the midline coelomic space that was not invaded by migrating primary mesenchyme.
- The notochord is important because it induces:
  formation of the head process,
  development of the nervous system, and
  formation of somites
- The notochord marks the future location of the vertebral column and the base of the cranium.
- The ultimate fate of the notochord is to become nucleus pulposus of intervertebral discs.

Note: The notochord develops from the *primitive node* located at the cranial end of the primitive streak. From the node, mesoderm-forming cells proliferate and migrate forward into the future head region where they become the rod-shaped notochord.



Note: Each organ system has a critical period during development when it is most sensitive to external agents (teratogens) that produce birth defects.

Early Formation of the Nervous System (Neurulation):

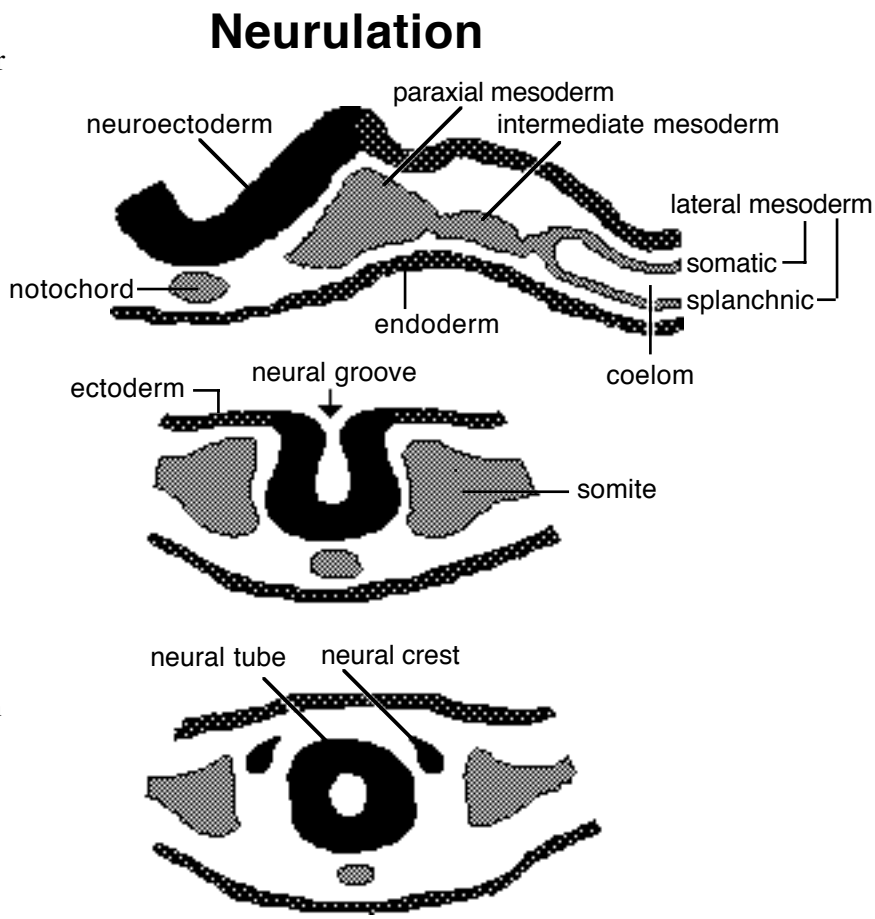
Neurulation refers to notochord-induced transformation of ectoderm into nervous tissue. The process begins during the third week in the region of the future brain and then progresses caudally into the region of the future spinal cord.

The neurulation process involves the following steps:

— ectodermal cells overlaying the notochord become tall columnar (neuroectoderm); they form a thickened area designated the *neural plate*. The other ectodermal epithelium is flattened.

— a *neural groove* is formed as edges of the neural plate become raised on each side of a midline depression. (Apical ends of individual neuroectodermal cells constrict.)

— a *neural tube* is then formed as the neural groove undergoes midline merger of its dorsal edges. The tube separates from non-neural ectoderm which unites dorsal to it. (Tube formation begins in the cranial cervical region of the central nervous system and progresses cranially and caudally until *anterior* and *posterior neuropores*, the last openings, finally close.)



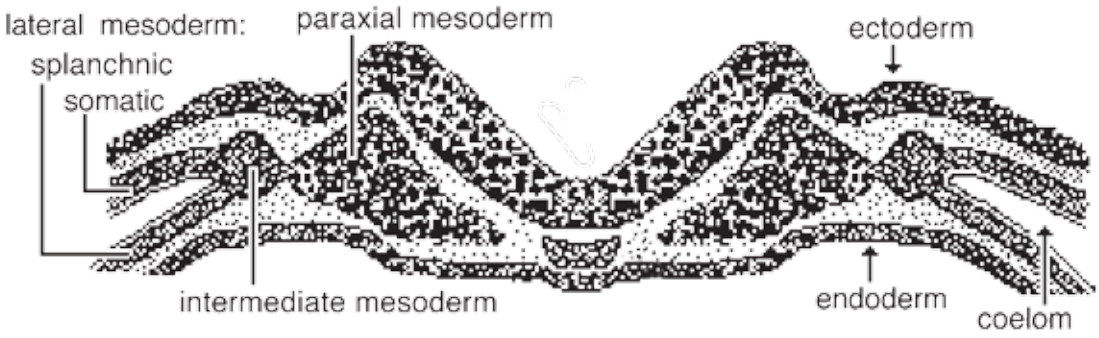
— bilaterally, where the neural groove is joined to non-neural ectoderm, cells detach as the neural groove closes; the cells proliferate and assume a position dorsolateral to the neural tube—forming *neural crest*.

NOTE:
Neural tube becomes the central nervous system, i.e., the brain and spinal cord.

Neural crest cells are remarkable for the range of structures they form. Some cells migrate dorsally and become pigment cells in skin. Other cells migrate ventrally and become neurons and glial cells of the peripheral nervous system, or adrenal medulla cells. In the head, neural crest forms mesenchyme (ectomesenchyme) which becomes meninges, bone, fascia, and teeth.

Somites:

- Mesoderm blocks located just lateral to the notochord, which induced somite development.
- A pair of somites develops for every vertebra, plus a half dozen somite pairs in the head.
- Number of somites in an embryo is indicative of age, individual somites develop chronologically, in craniocaudal order.



Somites develop as follows:

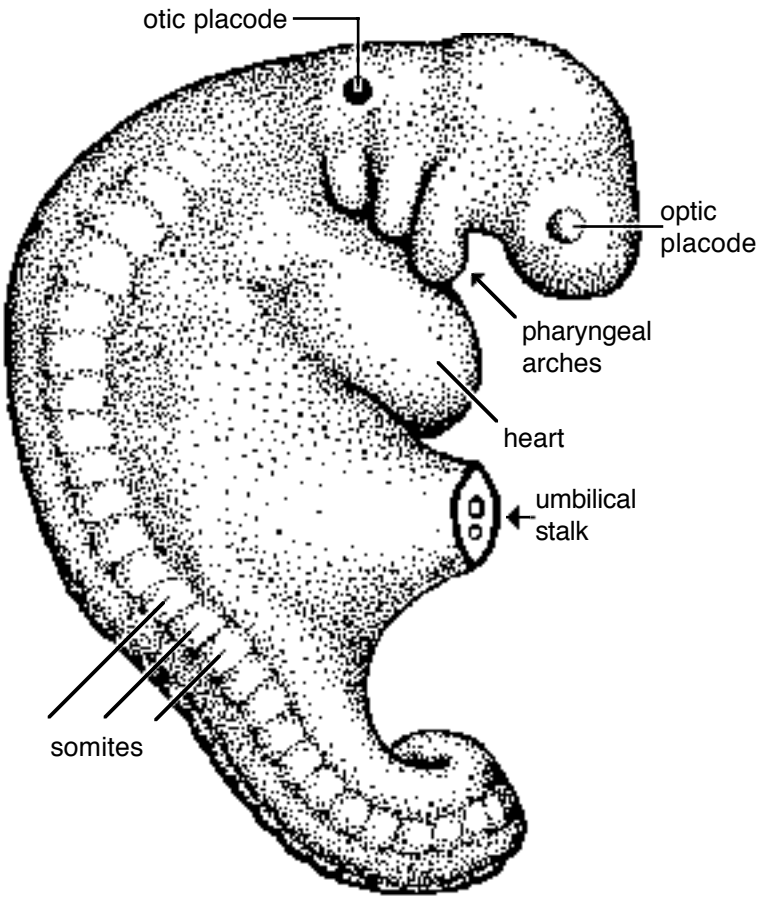
— mesoderm, designated ***paraxial mesoderm***, accumulates on each side of the notochord

— progressing from rostral to caudal over time, transverse fissures divide the paraxial mesoderm into blocks

— each block becomes a **somite** (epithelioid cells within a somite block re-orient 90°, from transverse to the notochord to longitudinal)

— head (occipital) somites develop from proliferation of local mesenchyme lateral to the cranial end of the notochord

— rostral to the notochord, mesenchyme forms less-developed somites, called ***somitomeres***; these migrate into pharyngeal arches and form muscles of the jaw, face, pharynx, & larynx.



NOTE:
Each somite differentiates into three regions:

Sclerotome (ventromedial region) gives rise to vertebrae, ribs, and endochondral bones at the base of the skull.

Dermatome (lateral region) gives rise to the dermis of skin

Myotome (intermediate region) gives rise to skeletal muscles of the body

Development of a Cylindrical Body:

The early embryo is flat, but the vertebrate body plan features a cylindrical theme—various cylindrical structures (derivatives of the gut, neural tube, notochord, etc.) enclosed within a cylindrical body. Transition from a flat embryo to a cylindrical one involves the following developments:

Head Process Formation:

• The cranial end of the embryo grows dorsally and forward so that it projects above the region originally in front of the embryo.

• The cylindrical head process elongates by additional growth from its base (located in front of the primitive node). Consequently, the most anterior part of the embryo is the oldest. The elongation incorporates the most anterior half-dozen somites into the future head.

• Within the head process, endoderm is reflected ventrally upon itself, forming a blind-ended *foregut* (future pharynx).

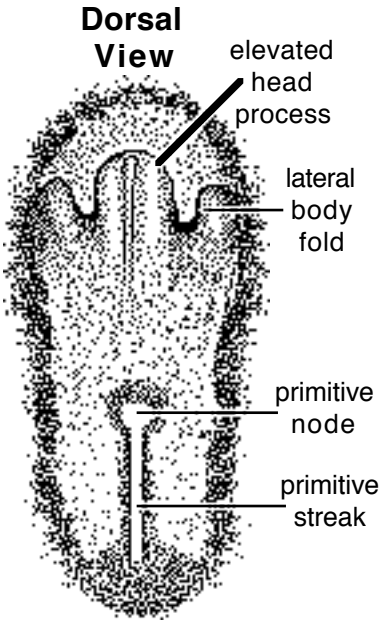
Tail Fold Formation:

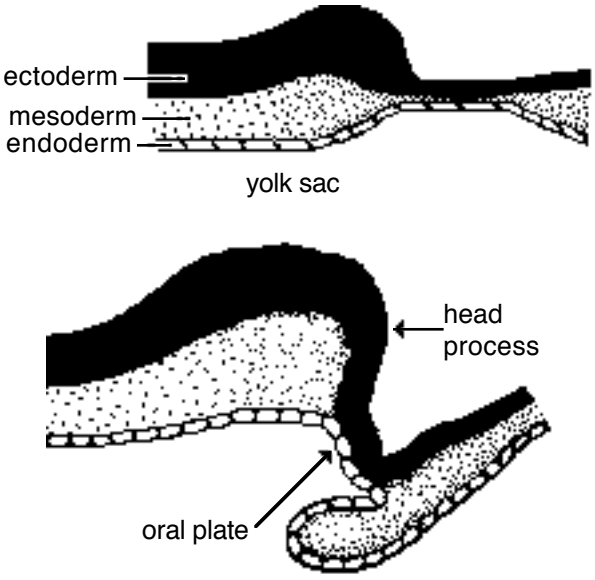
• At the caudal end of the embryo, a cylindrical tail fold is formed in a manner similar to that of the head process.

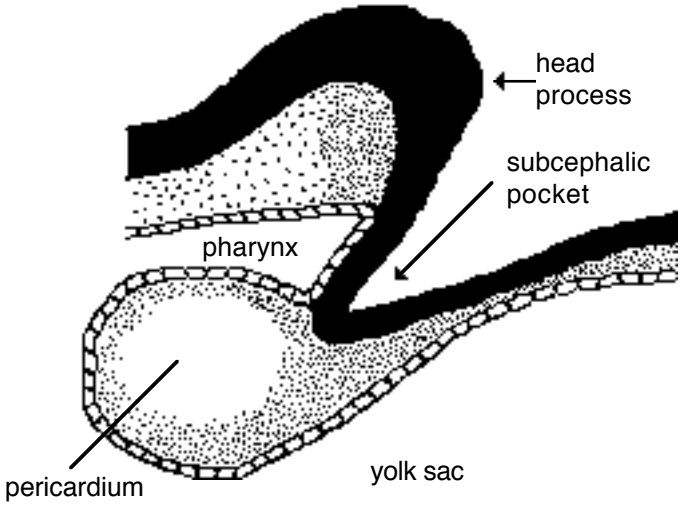
• Folded endoderm encloses a blind *hindgut* .

Lateral Body Folds:

• As the head process elongates upward & forward, a subcephalic pocket (space) is formed ventral to the head process, between the head process and extra-embryonic tissue. The bilateral margins of this pocket are lateral body folds—which constitute the continuity between the elevated embryo and the relatively flat extra-embryonic tissue.







• Similar folds exist caudally in association with the tail process.

• As the embryo grows and is elevated dorsally, lateral body folds adduct and join together ventrally, establishing a tubular embryo separated from flattened extra-embryonic tissue.

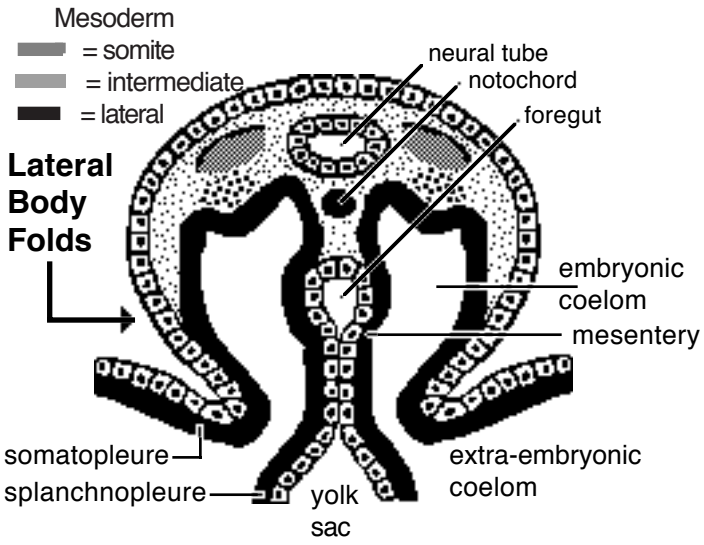
• Progressing caudally from the head process and cranially from the tail fold, ventral fusion of lateral body folds stops at the *umbilicus*—leaving a ventral opening in the body wall that allows vessels and the yolk sac and allantois to enter the embryo (and communicate with the gut).

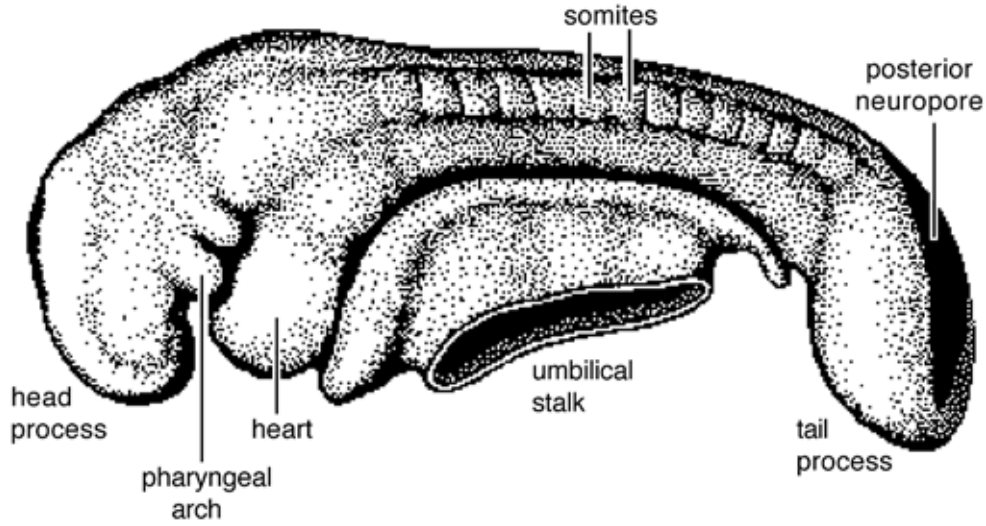
• Ventral fusion of lateral body folds distinguishes the embryo from extra-embryonic tissue (fetal membranes):

— embryonic coelom (future body cavities of the trunk) is distinguished from extra-embryonic coelom within fetal membranes.

— somatopleure (somatic mesoderm + ectoderm) that forms body wall is distinguished from that forming fetal membranes (chorion and amnion).

— splanchnopleure (splanchnic mesoderm + endoderm) merges bilaterally to form gut and mesentery, differentiated from extra-embryonic yolk sac (and allantois).

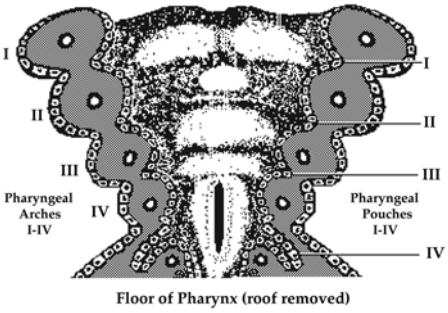




Pharyngeal Arches:

In the head region, dorso-ventral arches demarcated by grooves (clefts) appear. The arches are called ***pharyngeal arches*** and they are bounded internally by *pharyngeal pouches*.

Each arch contains a vessel (aortic arch). Within each arch, ectomesenchyme (derived from neural crest) gives rise to bone and fascia. Myotomes of somitomeres migrate to pharyngeal arches to provide skeletal musculature. Each arch is innervated by one cranial nerve.



Floor of Pharynx (roof removed)

Only the first three *pharyngeal arches* are externally evident in mammals. The first arch develops into upper and lower jaws and muscles of mastication. The second gives rise to hyoid bones and muscles of the face. The remaining pharyngeal arches form hyoid bones, larynx and associated muscles. Each arch is innervated by a particular cranial nerve.

The pharynx (foregut) develops five bilateral diverticula that internally demarcate the pharyngeal arches. These *pharyngeal pouches* develop into auditory tube, parathyroid glands, thymus, etc.

NOTE: In fish, five or six branchial [Gr. = gill] arches are well developed. Cells degenerate where branchial clefts and pharyngeal pouches meet so that the pharynx communicates with the outside (this occurs only temporarily between the first two arches in mammals). The first arch forms the jaw apparatus and the rest form gill arches separated by gill slits.

Flexures:

The tube-shaped embryo undergoes three flexures that make it C-shaped. The first occurs in the future midbrain region, the second in the future neck region, and the third occurs in the tail region.

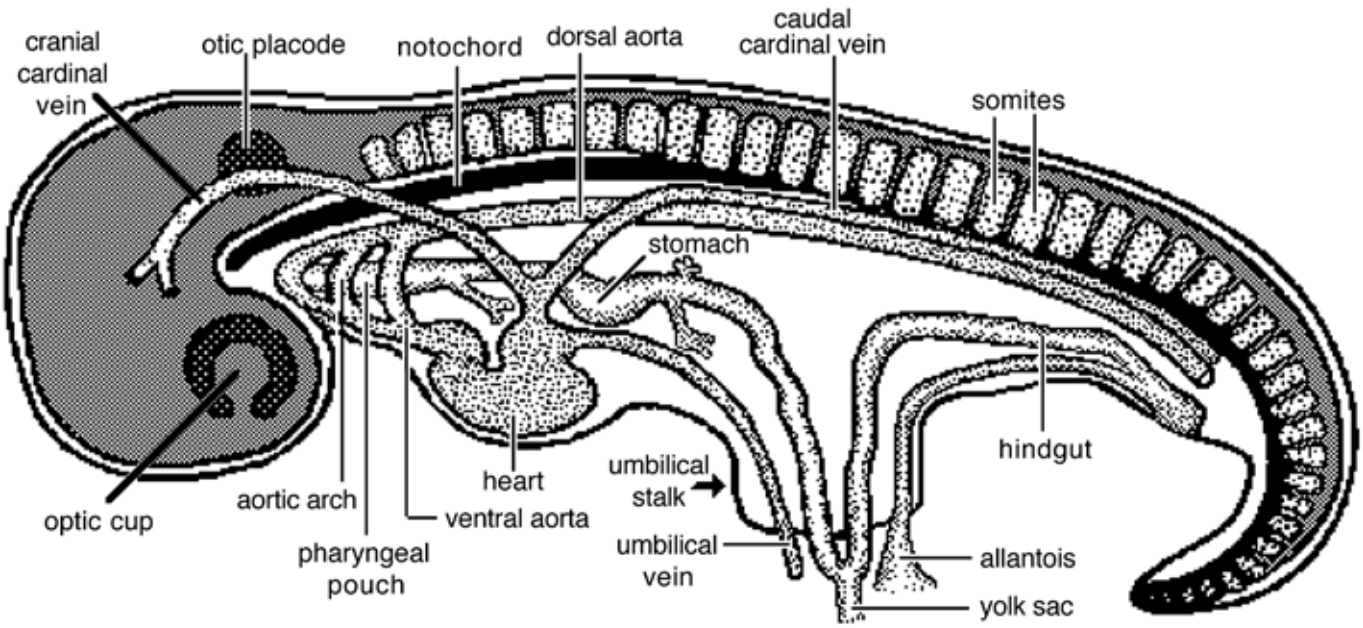
Cardiovascular system:

• The cardiovascular system develops early (in the third week after the start of the nervous system), as the embryo enlarges and diffusion alone becomes inadequate for tissue preservation.

• Angiogenesis (formation of blood vessels) begins in splanchnic mesoderm of the yolk sac, in the form of blood islands composed of mesenchyme and hemocytoblasts. The latter forms blood cells and the mesenchyme forms vesicles lined by endothelium. The vesicles coalesce to form vascular channels and then blood vessels (the latter are formed by budding, fusion, & enlargement).

• Vessels are formed first in extra-embryonic tissue: vitelline (yolk sac) and umbilical (allantoic) vessels appear first.

• Ventral to the pharynx, bilateral vessels merge to form a tubular heart; dorsal and ventral aortae are connected by aortic arches. Also, cranial and caudal cardinal veins return embryonic blood to the heart and umbilical veins return placental blood to the heart. None of these vessels will persist as such in the adult.



Placentation

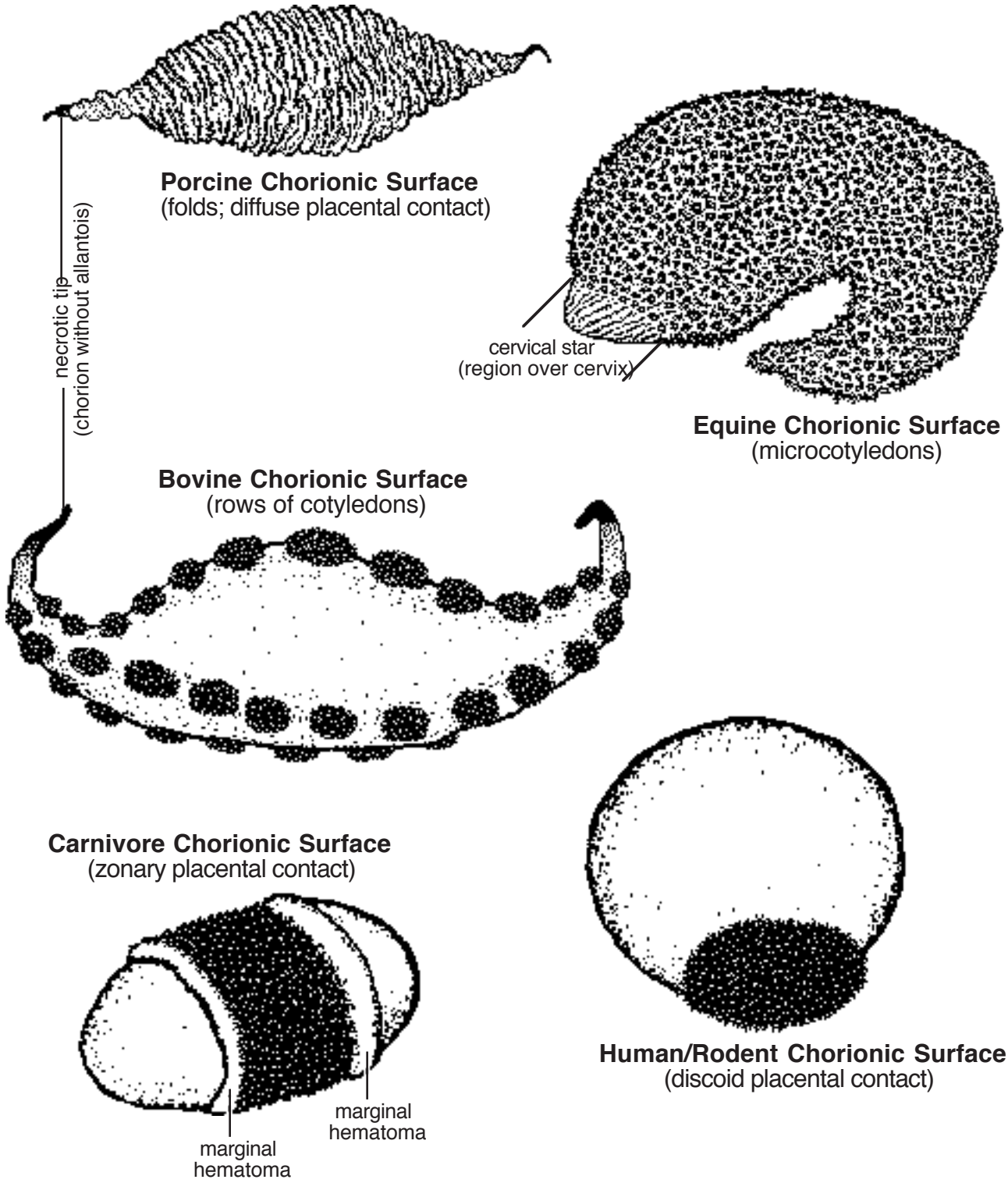
Placenta = region(s) of apposition between uterine lining and fetal membranes where metabolites are exchanged for sustaining pregnancy.

• Chorion forms the surface fetal membrane. Apposition areas (placental types) may be: *diffuse* (pig), *zonary* (carnivore), *discoid* (primates & rodents), or involve placentomes.

• A *placentome* is a discrete area of interdigitation between a maternal caruncle and a fetal cotyledon. Equine placentas are microcotyledonary (microplacentomes are distributed diffusely). Ruminant placentas consist of rows of relatively large placentomes.

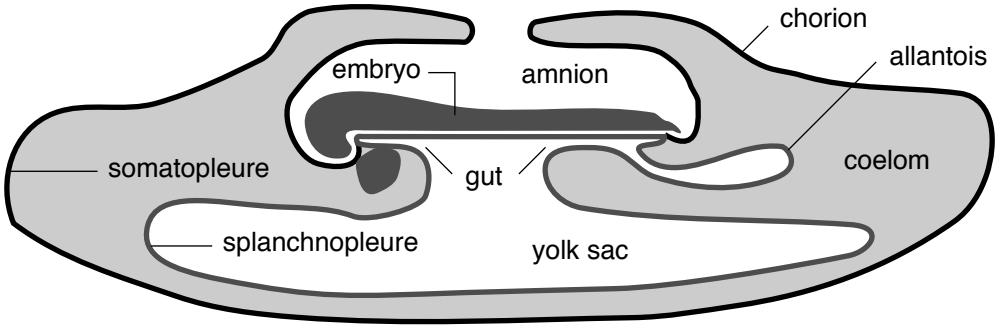
• Placentas (placentae) may also be classified according to the tissue layers separating fetal and maternal blood. Uterine epithelium, uterine connective tissue and uterine endothelium may be eroded, giving rise to four placental types: epitheliochorial (swine, equine, cattle); synepitheliochorial, formerly called syndesmochorial, (sheep, goats); endothelial chorial (carnivore); and hemochorial (primates & rodents).

Fetal Components of Placentae



Fetal membranes:

Four fetal membranes develop in a conceptus. Two arise from the trophoblast layer of the blastocyst (and are continuous with the somatopleure of the embryo). Two arise from the inner cell mass of the blastocyst (and are continuous with splanchnopleure of the embryo); these two splanchnopleure membranes are vascular. The four fetal membranes are:



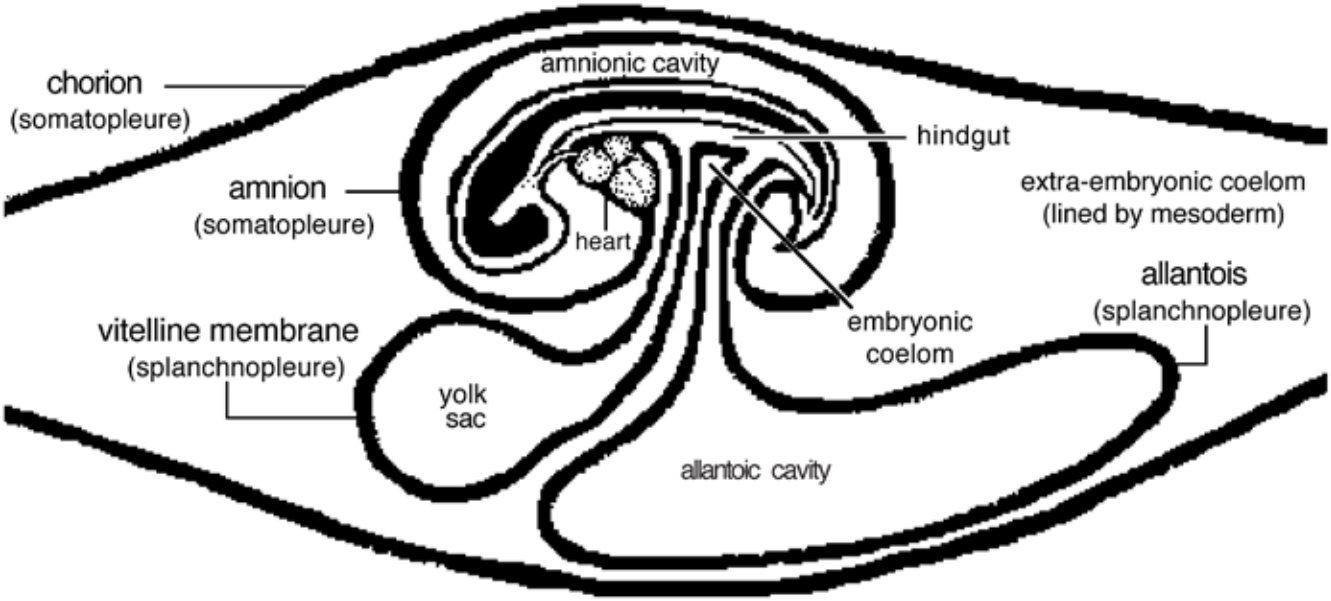
1. **Chorion** — forms the outer boundary of the entire conceptus (from trophoblast)

2. **Amnion** — encloses the embryo within a fluid-filled amnionic cavity; formed by folds of chorion in domestic mammals (in humans, amnion forms by cavitation deep to a persistent trophoblast).

3. **Allantois** — develops as an outgrowth of hindgut splanchnopleure (originates from inner cell mass). Allantois grows to fill the entire extra-embryonic coelom, with fluid-filled allantoic cavity in domestic mammals. The outer surface of allantois binds to the inner surface of chorion (and the outer surface of amnion). The allantois is highly vascular and provides the functional vessels of the placenta, via umbilical vessels.

4. **Yolk sac** — continuous with midgut splanchnopleure (develops early with hypoblast formation from inner cell mass). Supplied by vitelline vessels, it forms an early temporary placenta in the horse and dog. Yolk sac is most important in egg laying vertebrates.

Note: The term *conceptus* refers to the embryo or fetus plus its fetal membranes.



Implantation

The blastocyst is initially free in the uterine lumen (nourished by uterine glands). Implantation of the blastocyct is a gradual process, beginning with apposition, leading to adhesion (or invasion in the case of the human & Guinea Pig).

Approximate implantation times are: one week (human); two weeks (dog, cat, sheep), 3-5 weeks (cattle), 3-8 weeks horse; or delayed up to 4 mons (deer, bears).